

Abstract

Provided are methods of screening for the occurrence of gene silencing (e.g. post transcriptional gene silencing) in an organism (e.g. a plant or animal), which method comprises the steps of:

(i) obtaining a sample of material from said organism,  
(ii) producing a nucleic acid extract from said sample,  
(iii) analysing said extract such as to determine the presence or absence of short RNA molecules which are approximately 25 nucleotides in length (SRMs) in said nucleic extract,  
(iv) correlating the presence of said SRMs in the extract with the occurrence of gene silencing in said organism. The SRMs are preferably short anti-sense RNA molecules (SARMs). Also provided are associated methods for detecting the silencing of a target gene in an organism. Processes for isolating one or more RNA molecules, such as SARMs, which may be advantageously employed in the method, may include a purification step selected from (i) filtration; (ii) differential precipitation (iii) ion exchange chromatography, followed by separation the purified RNA molecules according to size by electrophoresis through 15% polyacrylamide gel containing 7M urea as a denaturant and TBE (0.5x) as a buffer, and blotting by electrophoresis. Also provided are processes for isolating silencing agents comprising SRMs; methods of selecting target regions of target genes for directed silencing; and methods of silencing target genes in organisms based on these. Silencing in a first organism may be achieved by generating in a second organism, SRMs which are introduced into the first organism such as to silence a target gene therein. Also provided are DNA constructs, host cells, plants and non-human mammals which comprise target genes which have been silenced in accordance with the methods herein.